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EXAMINER

KIM, ALEXANDER D

ART UNIT	PAPER NUMBER
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1656

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PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No. 10/812,315	Applicant(s) RIEPING, MECHTHILD	
	Examiner ALEXANDER D. KIM	Art Unit 1656	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

- 1) ☒ Responsive to communication(s) filed on 26 February 2009.
- 2a) ☐ This action is **FINAL**. 2b) ☒ This action is non-final.
- 3) ☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

- 4) ☒ Claim(s) 13-26 is/are pending in the application.
- 4a) Of the above claim(s) _____ is/are withdrawn from consideration.
- 5) ☐ Claim(s) _____ is/are allowed.
- 6) ☒ Claim(s) 13-26 is/are rejected.
- 7) ☐ Claim(s) _____ is/are objected to.
- 8) ☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

- 9) ☐ The specification is objected to by the Examiner.
- 10) ☐ The drawing(s) filed on _____ is/are: a) ☐ accepted or b) ☐ objected to by the Examiner.
Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).
Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).
- 11) ☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

- 12) ☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).
- a) ☐ All b) ☐ Some * c) ☐ None of:
1. ☐ Certified copies of the priority documents have been received.
 2. ☐ Certified copies of the priority documents have been received in Application No. _____.
 3. ☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

- | | |
|--|---|
| 1) <input checked="" type="checkbox"/> Notice of References Cited (PTO-892) | 4) <input type="checkbox"/> Interview Summary (PTO-413) |
| 2) <input type="checkbox"/> Notice of Draftsperson's Patent Drawing Review (PTO-948) | Paper No(s)/Mail Date. _____ |
| 3) <input checked="" type="checkbox"/> Information Disclosure Statement(s) (PTO/SB/08) | 5) <input type="checkbox"/> Notice of Informal Patent Application |
| Paper No(s)/Mail Date <u>03/04/2009</u> . | 6) <input type="checkbox"/> Other: _____ |

DETAILED ACTION

Application Status

1. In response to the previous Office action, a non-Final rejection (mailed on 10/31/2008), Applicants filed a response and amendment received on 02/26/2009. Said amendment cancelled Claims 1-12; amended Claims 13, 15-16, 21-23.

Claims 13-26 are pending in the instant Office action and will be examined herein.

It is noted that the instant office action is a non-final Office action in view of at least one new grounds of rejection.

Information Disclosure Statement

2. The information disclosure statement (IDS) filed on 03/04/2009 has been reviewed, and its reference has been considered. A copy of Form PTO/SB/08 is attached to the instant Office action.

Claim Objections

3. Claims 13-26 are objected to because of the following informalities:

The use of abbreviation "galP" in Claim 13 (Claims 14-22 dependent therefrom); abbreviation "PTS" in Claim 23 (Claims 24-26 dependent therefrom). Abbreviations, unless otherwise obvious and/or commonly used in the art, *e.g.*, "DNA", should not be recited in the claims without at least once reciting the entire phrase for which the

abbreviation is used in its first appearance in the claims. Appropriate correction is required.

Withdrawn-Claim Rejections - 35 USC § 112

4. The previous rejection of Claims 13 (Claims 14-22 dependent therefrom) and 23 (Claims 24-26 dependent therefrom) under of 35 U.S.C. 112, second paragraph, for Claims 13 (Claims 14-22 dependent therefrom) and Claim 23 (Claims 24-26 dependent therefrom) reciting limitations "fermenting a bacterium comprising an overexpressed endogenous DNA sequence encoding the galactose-proton symporter protein in said bacterium", "said bacterium is an Enterobacteriaceae family" and "said galactose-proton symporter protein comprises the amino acid sequence of SEQ ID NO: 4 and is encoded by the nucleotide sequence of SEQ ID NO: 3"; whereas the Enterobacteriaceae bacterium has to be E. coli because the nucleotide SEQ ID NO: 3 and the polypeptide of SEQ ID NO: 4 is the E. coli nucleotide or polypeptide (see instant specification page 10, lines 9-11); and as being unclear as to how any Enterobacteriaceae family other than E. coli can comprise an overexpressed endogenous DNA of SEQ ID NO: 3, or a DNA encoding SEQ ID NO: 4; is withdrawn by virtue of deleting the term "endogenous".

5. The previous rejection of Claims 21 and 22 under of 35 U.S.C. 112, second paragraph, for reciting the limitation "the thrABC operon" or "the tdh gene" wherein the recitation of "the" in front of the recited operon or gene as referring to the one specific

operon or gene; is withdrawn by virtue of applicant's amendment (i.e., amended to recite, e.g., "a thrABC operon" and "a tdh gene").

Claim Rejections - 35 USC § 112

The following is a quotation of the second paragraph of 35 U.S.C. 112:

The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

5. Claim 21 is rejected under of 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim 21 recites "using a strong promoter, and mutating the ribosome binding site". With respect to the limitation of "a strong promoter", the scope of those promoters that are considered to be "strong" is indefinite because the term "strong" is a relative term and neither the specification nor the claims provide a reference for determining whether or not the recited promoter is considered to be "strong". Moreover, it is unclear as to whether or not the limitation encompasses the naturally-occurring promoter, which, without a reference promoter as noted above, may be considered to be encompassed by the term "strong promoter".

Also, the limitation "the ribosome binding site" lacks antecedent basis in the claim because a ribosome binding site is not recited previously. The term "the" refers to a one particular ribosome binding site; and it is unclear what kind of change (or mutation) in any particular ribosome binding site would result in overexpression of the gene(s) recited in Claim 21.

Furthermore, it is noted that the claim requires overexpression of "one or more genes", where "using a strong promoter" and/or "mutating the ribosome binding site" may result in protein overexpression, but would not result in overexpression of one or more genes.

The following is a quotation of the first paragraph of 35 U.S.C. 112:

The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

6. Claim 21 is rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. The claim(s) contain subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention. This is a written description rejection.

According to MPEP 2163.II.A.1, in evaluating a claimed invention for adequate written description, the examiner should determine what the claim as a whole covers. "Claim construction is an essential part of the examination process. Each claim must be separately analyzed and given its broadest reasonable interpretation in light of and consistent with the written description. See, e.g., *In re Morris*, 127 F.3d 1048, 1053-54, 44 USPQ2d 1023, 1027 (Fed. Cir. 1997)."

CLAIM INTERPRETATION: Claim 21 limits the genus of microorganisms of the method of claim 13 to further overexpressing one or more of the genes as recited in the claim by (in relevant part) mutating the ribosome binding site, wherein the ribosome

binding site has been interpreted as a site on an mRNA expressed from the corresponding gene to which a ribosome binds.

MPEP 2163.II.A.2.(a).i) states, "Whether the specification shows that applicant was in possession of the claimed invention is not a single, simple determination, but rather is a factual determination reached by considering a number of factors. Factors to be considered in determining whether there is sufficient evidence of possession include the level of skill and knowledge in the art, partial structure, physical and/or chemical properties, functional characteristics alone or coupled with a known or disclosed correlation between structure and function, and the method of making the claimed invention".

The specification fails to disclose the reduction to practice of even a single representative species of the genus of microorganisms with a mutated ribosome binding site that results in overexpression of one or more of the genes as recited in the claims. There are no other drawings or structural formulas disclosed of a microorganism with a mutated ribosome binding site that results in overexpression of one or more of the genes as recited in the claims. The prior art does not teach any mutated ribosome binding site for recited genes. Thus, the specification and prior art do not teach any correlation between structure of any ribosome and function of being able to overexpress recited protein encoded by the gene. There is no evidence of record that the ribosome binding site(s) for expressed mRNAs corresponding to the recited genes were known in the art at the time of the invention. Moreover, there is no teaching or guidance in the specification or the prior art as to which nucleotide(s) of the mRNA corresponding to the

recited genes could be modified and result in overexpression of the gene. As such, the specification fails to satisfy the written description requirement of 35 U.S.C. 112, first paragraph. Because the Claim 21 encompasses very wide structural limitation of any mutation in any ribosome binding site for recited genes with no functional relationship to its structural changes in the genus of claim which would results in overexpression of said gene(s), the one skilled in the art would not be in possession of full scope of the claimed genus of the instant specification.

Withdrawn-Claim Rejections - 35 USC § 112

7. The previous rejection of Claim 23 (Claims 24-26 dependent therefrom) under 35 U.S.C. 112, first paragraph, **new matter**, for reciting "transports glucose by a PEP-dependent phosphotransferase (PTS) pathway", which is not supported by the original disclosure, is withdrawn by virtue of applicant's amendment.

8. The previous rejection of Claims 21-22 under 35 U.S.C. § 112, first paragraph, written description, wherein the breadth of Claim 21 includes a method comprising using an Enterobacteriaceae family having increased catalytic activity of enzymes from the thrABC operon according to the disclosure of the term "overexpression" on p. 7, line 1-5, is withdrawn by virtue of applicant's amendment (i.e., adding the limitation "increasing copy number" in claim 21; and adding "deleted" in Claim 22.

9. The previous rejection of Claims 21-22 under 35 U.S.C. § 112, first paragraph,

scope of enablement, because the specification, is withdrawn by virtue of applicant's amendment (i.e., adding the limitation "increasing copy number" in claim 21; and adding "deleted" in Claim 22.

Withdrawn-Claim Rejections - 35 USC § 102

10. The previous rejection of Claim 13-16, 18-20 under 35 U.S.C. 102(b) as being anticipated by Valle et al. (USPAP 2002/0155521 published on Oct. 24, 2002, as cited in the previous Office Action) as evidenced by Blattner et al. (1997, Science 277:1453-1474, as cited in the IDS), is withdrawn by virtue of applicant's amendment (i.e., deleting the term "operably linking said DNA to a promoter" and adding "changing a promoter normally found in a galP gene" wherein the reference of Valle et al. did not change a promoter of the galP gene).

11. The previous rejection of Claims 13-14 and 17-20 under 35 U.S.C. 102(a) as being anticipated by Hernandez-Montalvo et al. (2003 Sep. 20, Biotechnol Bioeng, Vol. 83, page 687-694, as cited previously on 12/13/2006) as evidenced by Blattner et al. (1997, Science 277:1453-1474, as cited in the IDS) and Lee et al. (2003, September, Journal of Bacteriology, vol. 185, p. 5442-5451, as cited previously on 12/13/2006), is withdrawn by virtue of applicant's amendment (i.e., reciting the correct coding sequence of SEQ ID NO: 4 which is residues 33-1427 of SEQ ID NO: 4; thus, it is fully supported by the instant foreign priority making the instant priority date to 4/1/2003).

Claim Rejections - 35 USC § 102

The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

A person shall be entitled to a patent unless –

(b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

12. Claims 13-20 and 23-24 are rejected under 35 U.S.C. 102(b) as being anticipated by **Martin et al.** (Biochem. J., 1995, Vol. 308, pages 261-268) as evidenced by **Roberts et al.** (Journal of Cellular Biochemistry, **1990**, Vol. 44, Issue S14E, on pages 1-30), "Biosynthesis of amino acids" (prepared by Bryant Miles, April 21, 2003), QIAGEN-Plasmid Resource Center (QIAGEN product description of Growth of bacterial cultures, printed 5/12/2009), and Berry et al. (TIBTECH, 1996, Vol. 14, pages 250-256).

Martin et al. teach cloning and overexpression of the galactose-H⁺ transport protein, GalP, of Escherichia coli; wherein the E. coli galP gene has 100 % identity to 33-1427 of SEQ ID NO: 3, and the encoded GalP protein has 100% identity to SEQ ID NO: 4 as evidenced by the sequence alignments below. Martin et al. teach growing the GalP protein producing E. coli JM1100 containing plasmid pPER3 containing galP overexpressing the GalP protein (see "Growth of the GalP-producing E. coli strain" on page 262, top of left column). Martin et al. teach the "yield of GalP in typical preparation was 40-60% of the inner membrane proteins" (see the end of "Quantification of overexpressed protein" on page 262, left column). Thus, the method step of Martin et al. meets the limitations of fermenting an Enterobacteriaceae family overexpressed DNA sequence encoding the galactose-protein symporter protein (i.e., SEQ ID NO: 4

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encoded by the nucleic acid of 33-1472 of SEQ ID NO: 3 which is the E. coli galP gene) wherein the overexpression is achieved by changing a promoter normally found in a galP gene. The E. coli of Martin et al. overexpressing GalP protein inherently produces amino acids (e.g., L-threonine, L-isoleucine, L-valine, L-methionine, L-homoserine and L-lysine) from a glucose, when the cell were grown in a media, as evidenced by (and it is well known in the art) the reference of "Biosynthesis of amino acids", which summarizes the biosynthesis of amino acids and teaches "bacteria can synthesize all 20 of the amino acids" (see bottom of page 1) and "the carbon skeletons comes from intermediates of glycolysis, the pentose phosphate pathway and the citric acid cycle" (see middle of page 2) wherein said glycolysis starts with a glucose molecule. Thus, the amino acid produced in the E. coli of Martin et al. is produced from glucose and the method of growing E. coli JM1100 overexpressing GalP protein meets all limitations of **Claims 13-16 and 18**, wherein an L-amino acid becomes enriched in a fermentation medium as the total contents of the E. coli (which contains all 20 L-amino acids) increases. Martin et al. teach isolation of the overexpressed GalP protein by French press and centrifugation (see middle of left column, page 262); thus, meeting the limitation of **Claims 19-20**.

The pPER3 vector is derived from pBR322 as evidenced by the reference of Roberts et al. (Abstract N109 at p. 20, top); wherein the pBR322 has a copy number of 15-20 as evidenced by the reference of Qiagen; thus, the method of Martin et al. meets the step of increasing copy number of said DNA in **Claim 17**.

The instant claim 23 has additional limitation of said Enterobacteriaceae bacterium comprises PTS enzymes. The instant PTS enzymes in a method of Claim 23 read on any PTS enzymes which exist in E. coli as evidenced by Berry et al. who disclose "The PTS is the major consumer of PEP in E. coli growing on glucose" (see Berry et al., lines 31-32, right column, page 254). Thus, in addition to the reasons above, the E. coli of Martin et al. has many PTS enzymes inherently and meets the limitation of method steps of **Claim 23-24**.

Withdrawn-Claim Rejections - 35 USC § 103

13. The previous rejection of Claim 21 under 35 U.S.C. 103(a) as being unpatentable over Valle et al. (USPAP 2002/0155521 published on Oct. 24, 2002, as cited in the previous Office Action) in view of Debabov et al. (USP 6,132,999 published on Oct. 17, 2000, as cited in the previous Office Action) is withdrawn by virtue of applicant's amendment overcoming the 35 USC 102 rejection as shown above.

14. The previous rejection of Claim 22 under 35 U.S.C. 103(a) as being unpatentable over Valle et al. (USPAP 2002/0155521 published on Oct. 24, 2002) in view of Debabov et al. (USP 5,705,371 published on Jan. 6, 1998), is withdrawn by virtue of applicant's amendment overcoming the 35 USC 102 rejection as shown above.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

(a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negated by the manner in which the invention was made.

15. Claims 13-20 and 23-26 are rejected under 35 U.S.C. 103(a) as being unpatentable over Debabov et al. (US Patent 5,175,107 issued on Dec. 29, 1992) in view of Venter et al. (Biochem. J., 2002, Vol. 363, pages 243-252), Flores et al. (Metabolic Engineering, 2002, Vol. 4, pages 124-137) and Berry et al. (TIBTECH, 1996, Vol. 14, pages 250-256) and as evidenced by Martin et al. (Biochem. J., 1995, Vol. 308, pages 261-268), and "Biosynthesis of amino acids" (prepared by Bryant Miles, April 21, 2003).

Debabov et al. (1992) teach teaches a method of making L-threonine using the E. coli BKIM B-3996 (see Example 1 and 2 in §4-5) containing a recombinant plasmid pVIC40 (see Abstract). Debabov et al. (1992) teach a method step of growing said E. coli in a medium having 4% by mass (see bottom of §4) and teach a method of determining the concentration of the L-threonine as Debabov et al. disclose the fermentation resulted in a total of 85g/l L-threonine. Debabov et al. also teach many carbon sources for a fermentation which "Exhibits good growth on saccharose, glucose, fructose, lactose, mannose, galactose, xylose, glycerol, mannitol" (see §3, lines 52-54).

The difference between Debabov et al. (1992) and the claimed invention is that the *E. coli* BKIM B-3996 of Debabov (1992) does not overexpress the *E. coli* galP gene (i.e., residues 33-1427 of SEQ ID NO: 3 encoding the SEQ ID NO: 4 protein).

Venter et al. teach the cloning and expression of *E. coli* GalP (SEQ ID NO: 4) encoded by the galP gene which were known to be the residues 33-1427 of SEQ ID NO: 3 as shown in the second (as early as 1980 by Smith et al.) and third (Blattner et al. 1997) sequence alignments at the end of the instant office action.

Flores et al. teach "that GalP can transport, in addition to galactose, other sugars such as glucose", see left column, lines 14-15, page 125, and further teaches that a functional galP gene on the chromosome is required for rapid growth on glucose and teach that it can direct more carbon flux into intermediates or final products of the aromatic pathway, compared to isogenic PTS⁺ strains (see left column, lines 18-20, page 125). Flores acknowledges the concept of metabolic engineering of *E. coli* for the improvement of industrial strains, including metabolic engineering studies on the main central metabolic sections that are active during growth on glucose as the sole carbon source, including overexpression of a gene encoding a protein involved in metabolic flux (see bottom of left column to right column on p. 124).

Berry et al. teach that influx of Glucose feeds into the glycolysis and the TCA cycles in Figure 4 on page 254 which produces intermediate compounds that are used as precursors of all amino acid biosynthesis; which is well known in the art and summarized by the disclosure of the evidentiary reference of "Biosynthesis of amino acids", which discloses that "bacteria can synthesize all 20 of the amino acids" (see

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bottom of page 1) and "the carbon skeletons comes from intermediates of glycolysis, the pentose phosphate pathway and the citric acid cycle" (see middle of page 2) wherein said precursor of amino acid biosynthesis starts with a glucose molecule.

Therefore, it would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to practice the method of Debabov et al. (1992) using the *E. coli* of Debabov et al. overexpressing the galP gene of Venter et al. where overexpression is achieved by transforming the *E. coli* with a vector encoding galP with a reasonable expectation of success because the GalP gene increases the carbon flux into the cell by transporting various sugar including glucose and it can be very effective transporting the sugar molecules specially when it can be expressed as high as 40 to 60 % of total membrane as shown by Martin et al. (see the end of "Quantification of overexpressed protein" on page 262, left column); wherein the increase in carbon source of sugar molecules increases the precursor for the biosynthesis of amino acids (L-threonine, for example). Debabov et al. (1992) teach the usefulness of L-threonine which is "known to be an essential amino acid applicable as the component of diverse nutritive mixture of medical use", "an additive to animals' fodder, as well as reagent for the pharmaceutical and chemical industries and as a growth factor for microorganisms producing some other amino acids, such as L-lysine and L-homoserine" (see §1, lines 12-18). Also, Flores et al. teach the PTS system (i.e., PEP:carbohydrate phosphotransferase system) is the major PEP-utilizing system wherein the PEP is major precursor of biosynthesis of amino acids (as supported by the glycolysis and the TCA cycles in Figure 4 on page 254 of Berry et al.) and "the usage of a different system to

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internalize and phosphorylate glucose, would lead to an increase in the amount of PEP" so that PEP would be used in the biosynthesis of many compounds as well as increase in feeding carbon source into a TCA cycle which in turn increase in amino acid biosynthesis such as L-threonine. Thus, one skilled in the art would recognize the GalP transport system as one of said different system as indicated by Flores et al. to internalize glucose besides the PTS system. One skilled in the art would be motivated to overexpress the E. coli GalP protein in a L-threonine producing E. coli (BKIM B-3996, for example) for increase glucose influx in addition to the central glucose influx PEP:carbohydrate phosphotransferase system (PTS) which would increase overall carbon source required for amino acids production which is advantageous in an industry for increasing a cost efficiency which is useful in the amino acid producing industry. Thus, the claimed invention as a whole was *prima facie* obvious over the combined teachings of the prior art.

16. Claim 21 is rejected under 35 U.S.C. 103(a) as being unpatentable over Debabov et al. (1992) in view of Venter et al., Flores et al., and Berry et al. and as evidenced by Martin et al., and "Biosynthesis of amino acids" as applied to claims 13-20 and 23-26 above, and further in view of Debabov et al. (USP 6,132,999 published on Oct. 17, 2000, as cited in the previous non-final office action mailed on 10/31/2008).

The teachings of Debabov et al. (1992), Venter et al., Flores et al., and Berry et al. are set forth above. The combination of Debabov et al. (1992), Venter et al., Flores et al., and Berry et al. does not teach or suggest additionally overexpressing a gene as recited in claim 21.

Debabov et al. (2000) teach a process of improved amino acid production by transforming an *E. coli* with an expression vector comprising a threonine operon (thrABC), which overexpresses the thrABC gene product. Debabov et al. (2000) teach a process of making L-threonine by using *E. coli* BKIIM B-5318 in Example 1. The *E. coli* BKIIM B-5318 has "plasmid pPRT614, which has threonine biosynthesis genes (thrA, B, and C)" as disclosed in the Abstract.

At the time of the invention, it would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to practice the method of Debabov et al. (1992) using the *E. coli* of Debabov et al. (1992) overexpressing the galP gene of Venter et al and further overexpressing the thrABC operon of Debabov et al. (2000), where overexpression is achieved by transforming the *E. coli* with a vector encoding galP and thrABC of Debabov et al. (2000) with a reasonable expectation of success because molecular biology techniques for cloning and expressing multiple genes using a plasmid vector are well known in the art, and in view of teachings of thrABC operon by Debabov et al. (1992) and the teaching of *E. coli* galP gene and its function by Venter et al. (2002), Flores et al. (2002), Berry et al. (1996). The motivation to do so is provided by Debabov et al. (2000) et al. who teaches the usefulness of cost-effective and efficient biosynthetic production of compounds including the L-Thr and that overexpression of thrABC operon results in enhanced L-Thr production as taught by Debabov et al. (2000). Thus, the claimed invention as a whole was *prima facie* obvious over the combined teachings of the prior art.

17. Claim 22 is rejected under 35 U.S.C. 103(a) as being unpatentable over Debabov et al. (1992) in view of Venter et al., Flores et al., and Berry et al. and as evidenced by Martin et al., and "Biosynthesis of amino acids" as applied to claims 13-20 and 23-26 above, and further in view of Debabov et al. (USP 5,705,371 published on Jan. 6, 1998, as cited in the previous non-final office action mailed on 10/31/2008).

The teachings of Debabov et al. (1992), Venter et al., Flores et al., and Berry et al. are set forth above. The combination of Debabov et al. (1992), Venter et al., Flores et al., and Berry et al. does not teach or suggest deletion of a gene as recited in claim 22.

Debabov et al. (1998) teach a process of making L-threonine by attenuation of the *tdh* gene encoding a threonine dehydrogenase "engaged in degradation of L-threonine" (see column 2, lines 58-59). Debabov et al. (1998) teach "E. coli strain VNIIgenetika 472T23" having "insertion of transposon Tn5 into gene *tdh* " is "devoid completely of activity" of a threonine dehydrogenase (see column 2, line 53-59).

At the time of the invention, it would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to practice the method of Debabov et al. (1992) using the *E. coli* of Debabov et al. (1992) overexpressing the *galP* gene of Venter et al with a deletion of a *tdh* gene as taught by Debabov et al. (1998), where overexpression is achieved by transforming the *E. coli* with a vector encoding *galP* with a reasonable expectation of success because molecular biology techniques for cloning and expressing multiple genes using a plasmid vector as well as the deletion of known gene sequences are well known in the art in view of teachings of deletion of *tdh* gene by

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inserting transposon by Debabov et al. (1998) and the teaching of E. coli galP gene and its function by Venter et al. (2002), Flores et al. (2002), Berry et al. (1996). The motivation to do so is provided by Debabov et al. (1998) who teach increasing the production of L-Thr production could be enhanced by deleting tdh gene encoding a polypeptide that degrades L-Thr. One skilled in the art would be motivated to delete the tdh gene for usefulness, cost-effective and efficient biosynthetic production of compounds including the L-Thr by deleting the tdh gene as taught by Debabov et al. (1998). Thus, the claimed invention as a whole was *prima facie* obvious over the combined teachings of the prior art.

Conclusion

18. Any inquiry concerning this communication or earlier communications from the examiner should be directed to Alexander D. Kim whose telephone number is (571) 272-5266. The examiner can normally be reached on 10AM-6:30PM.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Andrew Wang can be reached on (571) 272-0811. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see <http://pair-direct.uspto.gov>. Should

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you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

/Alexander D Kim/
Examiner, Art Unit 1656

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Sequence Alignments

10/812315
 RESULT 2
 GALP_ECOLI
 ID GALP_ECOLI STANDARD; PRT; 464 AA.
 AC P0AEP1; P37021;
 DT 20-DEC-2005, integrated into UniProtKB/Swiss-Prot.
 DT 20-DEC-2005, sequence version 1.
 DT 07-MAR-2006, entry version 5.
 DE Galactose-proton symporter (Galactose transporter).
 GN Name=galP; OrderedLocusNames=b2943;
 OS Escherichia coli.
 OC Bacteria; Proteobacteria; Gammaproteobacteria; Enterobacteriales;
 OC Enterobacteriaceae; Escherichia.
 OX NCBI_TaxID=562;
 RN [1]
 RP NUCLEOTIDE SEQUENCE [GENOMIC DNA], AND CHARACTERIZATION.
 RA Roberts P.E.;
 RL Thesis (1992), University of Cambridge, United Kingdom.
 RN [2]
 RP NUCLEOTIDE SEQUENCE [LARGE SCALE GENOMIC DNA].
 RC STRAIN=K12 / MG1655;
 RX MEDLINE=97426617; PubMed=9278503; DOI=10.1126/science.277.5331.1453;
 RA Blattner F.R., Plunkett G. III, Bloch C.A., Perna N.T., Burland V.,
 RA Riley M., Collado-Vides J., Glasner J.D., Rode C.K., Mayhew G.F.,
 RA Gregor J., Davis N.W., Kirkpatrick H.A., Goeden M.A., Rose D.J.,
 RA Mau B., Shao Y.;
 RT "The complete genome sequence of Escherichia coli K-12.";
 RL Science 277:1453-1474(1997).
 RN [3]
 RP SUBCELLULAR LOCATION.
 RC STRAIN=K12 / MG1655;
 RX PubMed=15919996; DOI=10.1126/science.1109730;
 RA Daley D.O., Rapp M., Granseth E., Melen K., Drew D., von Heijne G.;
 RT "Global topology analysis of the Escherichia coli inner membrane
 RT proteome.";
 RL Science 308:1321-1323(2005).
 CC -!- FUNCTION: Uptake of galactose across the boundary membrane with
 CC the concomitant transport of protons into the cell (symport
 CC system).
 CC -!- SUBCELLULAR LOCATION: Bacterial cell inner membrane; multi-pass
 CC membrane protein.
 CC -!- SIMILARITY: Belongs to the major facilitator superfamily. Sugar
 CC transporter family.
 CC -----
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 CC -----
 DR EMBL; U28377; AAA69110.1; -; Genomic_DNA.
 DR EMBL; U00096; AAC75980.1; -; Genomic_DNA.
 DR PIR; F65079; F65079.
 DR GenomeReviews; U00096_GR; b2943.
 DR EchoBASE; EB2068; -.
 DR EcoGene; EG12148; galP.
 DR BioCyc; EcoCyc:GALP-MONOMER; -.
 DR LinkHub; P37021; -.
 DR PROSITE; PS50850; MFS; 1.
 DR PROSITE; PS00216; SUGAR_TRANSPORT_1; 1.
 DR PROSITE; PS00217; SUGAR_TRANSPORT_2; 1.
 KW Complete proteome; Inner membrane; Membrane; Sugar transport; Symport;

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KW	Transmembrane; Transport.			
FT	CHAIN	1	464	Galactose-proton symporter. /FTid=PRQ_0000050292.
FT	TOPO_DOM	1	15	Cytoplasmic (Potential).
FT	TRANSMEM	16	36	1 (Potential).
FT	TOPO_DOM	37	56	Periplasmic (Potential).
FT	TRANSMEM	57	77	2 (Potential).
FT	TOPO_DOM	78	84	Cytoplasmic (Potential).
FT	TRANSMEM	85	105	3 (Potential).
FT	TOPO_DOM	106	112	Periplasmic (Potential).
FT	TRANSMEM	113	133	4 (Potential).
FT	TOPO_DOM	134	139	Cytoplasmic (Potential).
FT	TRANSMEM	140	160	5 (Potential).
FT	TOPO_DOM	161	171	Periplasmic (Potential).
FT	TRANSMEM	172	192	6 (Potential).
FT	TOPO_DOM	193	250	Cytoplasmic (Potential).
FT	TRANSMEM	251	271	7 (Potential).
FT	TOPO_DOM	272	290	Periplasmic (Potential).
FT	TRANSMEM	291	311	8 (Potential).
FT	TOPO_DOM	312	321	Cytoplasmic (Potential).
FT	TRANSMEM	322	342	9 (Potential).
FT	TOPO_DOM	343	351	Periplasmic (Potential).
FT	TRANSMEM	352	372	10 (Potential).
FT	TOPO_DOM	373	394	Cytoplasmic (Potential).
FT	TRANSMEM	395	415	11 (Potential).
FT	TOPO_DOM	416	416	Periplasmic (Potential).
FT	TRANSMEM	417	437	12 (Potential).
FT	TOPO_DOM	438	464	Cytoplasmic (Potential).
SO	SEQUENCE	464 AA;	50983 MW;	07E08935BD8E3F8E CRC64;

Query Match 100.0%; Score 2359; DB 1; Length 464;
Best Local Similarity 100.0%; Pred. No. 5.1e-149;
Matches 464; Conservative 0; Mismatches 0; Indels 0; Gaps 0;

Qy	1	MPDAKKQGRSNKAMTFFVCFLAALAGLLFGLDIGVIAGALPFIADefQITshtQEWVvSS	60
Db	1	MPDAKKQGRSNKAMTFFVCFLAALAGLLFGLDIGVIAGALPFIADefQITshtQEWVvSS	60
Qy	61	MMFGAAVGAVGSGWLSFKLGRKKSLMIGAILFVAGSLFSAAAPNVEVLIISRVLLGLAVG	120
Db	61	MMFGAAVGAVGSGWLSFKLGRKKSLMIGAILFVAGSLFSAAAPNVEVLIISRVLLGLAVG	120
Qy	121	VASYTAPLYLSEIAPEKIRGSMISMYQLMITIGILGAYLSDTAFsYtGAWRWMLGvIIIP	180
Db	121	VASYTAPLYLSEIAPEKIRGSMISMYQLMITIGILGAYLSDTAFsYtGAWRWMLGvIIIP	180
Qy	181	AILLIGVFFFLPDSPRWFaAKRRFvDAERVLLRLRDTsAEAKRELDEIReSLQVKQSGWA	240
Db	181	AILLIGVFFFLPDSPRWFaAKRRFvDAERVLLRLRDTsAEAKRELDEIReSLQVKQSGWA	240
Qy	241	LFKENSfRRaVFLGVLLQVMQqFTGMNVIMyYAPKIFeLAGYtNTtEQmWGTvIVGLTN	300
Db	241	LFKENSfRRaVFLGVLLQVMQqFTGMNVIMyYAPKIFeLAGYtNTtEQmWGTvIVGLTN	300
Qy	301	VlATfIAIGLVDRWGRKPTLTlGLFLVMAAGMGVLGTmmHIGIHsPSAQYfAIAMLLMFIV	360
Db	301	VlATfIAIGLVDRWGRKPTLTlGLFLVMAAGMGVLGTmmHIGIHsPSAQYfAIAMLLMFIV	360
Qy	361	GfAMsAGPLiWVLCSEIQPLKGRDFGItCSTATNWIANMIVGATfLTmLNTlGNANtFWV	420

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Db 361 GFAMSAGPLIWVLCSEIQPLKGRDFGITCSTATNWIANMIVGATFLTMLNTLGNANTFWV 420

QY 421 YAALNVLFILLTLWLVPETKHVSLEHIERNLMKGRKLREIGAH 464
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Db 421 YAALNVLFILLTLWLVPETKHVSLEHIERNLMKGRKLREIGAH 464

RESULT 4

Q2M9Q0_ECOLI

ID Q2M9Q0_ECOLI PRELIMINARY; PRT; 464 AA.

AC Q2M9Q0;

DT 21-FEB-2006, integrated into UniProtKB/TrEMBL.

DT 21-FEB-2006, sequence version 1.

DT 07-MAR-2006, entry version 2.

DE D-galactose transporter.

GN Name=galP;

OS Escherichia coli W3110.

OC Bacteria; Proteobacteria; Gammaproteobacteria; Enterobacteriales;

OC Enterobacteriaceae; Escherichia.

OX NCBI_TaxID=316407;

RN [1]

RP NUCLEOTIDE SEQUENCE.

RC STRAIN=K-12;

RX MEDLINE=81053692; PubMed=6159575;

RA **Smith** D.R., Calvo J.M.;

RT "Nucleotide sequence of the E coli gene coding for dihydrofolate

RT reductase.";

RL Nucleic Acids Res. 8:2255-2274(1980).

RN [2]

RP NUCLEOTIDE SEQUENCE.

RC STRAIN=K-12;

RA Sekiguchi T., Ortega-Cesena J., Nosoh Y., Ohashi S., Tsuda K.,

RA Kanaya S.;

RT "DNA and amino-acid sequences of 3-isopropylmalate dehydrogenase of

RT Bacillus coagulans. Comparison with the enzymes of Saccharomyces

RT cerevisiae and Thermus thermophilus.";

RL Biochim. Biophys. Acta 867:36-44(1986).

RN [3]

RP NUCLEOTIDE SEQUENCE.

RC STRAIN=K-12;

RA Chen H., Sun Y., Stark T., Beattie W., Moses R.E.;

RT "Nucleotide sequence and deletion analysis of the polB gene of

RT Escherichia coli.";

RL DNA Cell Biol. 9:613-635(1990).

RN [4]

RP NUCLEOTIDE SEQUENCE.

RC STRAIN=K-12;

RA Smallshaw J.E., Kelln R.A.;

RT "Cloning, nucleotide sequence and expression of the Escherichia coli

RT K-12 pyrH gene encoding UMP kinase.";

RL Genetics (Life Sci. Adv.) 11:59-65(1992).

RN [5]

RP NUCLEOTIDE SEQUENCE.

RC STRAIN=K-12;

RA Hayashi K., Morooka N., Yamamoto Y., Fujita K., Isono K., Choi S.,

RA Ohtsubo E., Baba T., Wanner B.L., Mori H., Horiuchi T.;

RT "Highly accurate genome sequences of Escherichia coli K-12 strains

RT MG1655 and W3110.";

RL Mol. Syst. Biol. 0:0-0(2006).

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RN [6]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX PubMed=16397293; DOI=10.1093/nar/gkj150;
RA Riley M., Abe T., Arnaud M.B., Berlyn M.K., Blattner F.R.,
RA Chaudhuri R.R., Glasner J.D., Horiuchi T., Keseler I.M., Kosuge T.,
RA Mori H., Perna N.T., Plunkett G. III, Rudd K.E., Serres M.H.,
RA Thomas G.H., Thomson N.R., Wishart D., Wanner B.L.;
RT "Escherichia coli K-12: a cooperatively developed annotation snapshot-
RT -2005.";
RL Nucleic Acids Res. 34:1-9(2006).
RN [7]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=97349980; PubMed=9205837; DOI=10.1093/dnares/4.2.91;
RA Yamamoto Y., Aiba H., Baba T., Hayashi K., Inada T., Isono K.,
RA Itoh T., Kimura S., Kitagawa M., Makino K., Miki T., Mitsuhashi N.,
RA Mizobuchi K., Mori H., Nakade S., Nakamura Y., Nashimoto H.,
RA Oshima T., Oyama S., Saito N., Sampei G., Satoh Y., Sivasundaram S.,
RA Tagami H., Takahashi H., Takeda J., Takemoto K., Uehara K., Wada C.,
RA Yamagata S., Horiuchi T.;
RT "Construction of a contiguous 874-kb sequence of the Escherichia coli
RT -K12 genome corresponding to 50.0-68.8 min on the linkage map and
RT analysis of its sequence features.";
RL DNA Res. 4:91-113(1997).
RN [8]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=97251358; PubMed=9097040; DOI=10.1093/dnares/3.6.379;
RA Itoh T., Aiba H., Baba T., Fujita K., Hayashi K., Inada T., Isono K.,
RA Kasai H., Kimura S., Kitakawa M., Kitagawa M., Makino K., Miki T.,
RA Mizobuchi K., Mori H., Mori T., Motomura K., Nakade S., Nakamura Y.,
RA Nashimoto H., Nishio Y., Oshima T., Saito N., Sampei G., Seki Y.,
RA Sivasundaram S., Tagami H., Takeda J., Takemoto K., Wada C.,
RA Yamamoto Y., Horiuchi T.;
RT "A 460-kb DNA sequence of the Escherichia coli K-12 genome
RT corresponding to the 40.1-50.0 min region on the linkage map.";
RL DNA Res. 3:379-392(1996).
RN [9]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=97251357; PubMed=9097039; DOI=10.1093/dnares/3.6.363;
RA Aiba H., Baba T., Fujita K., Hayashi K., Inada T., Isono K., Itoh T.,
RA Kasai H., Kashimoto K., Kimura S., Kitakawa M., Kitagawa M.,
RA Makino K., Miki T., Mizobuchi K., Mori H., Mori T., Motomura K.,
RA Nakade S., Nakamura Y., Nashimoto H., Nishio Y., Oshima T., Saito N.,
RA Sampei G., Seki Y., Sivasundaram S., Tagami H., Takeda J.,
RA Takemoto K., Takeuchi Y., Wada C., Yamamoto Y., Horiuchi T.;
RT "A 570-kb DNA sequence of the Escherichia coli K-12 genome
RT corresponding to the 28.0-40.1 min region on the linkage map.";
RL DNA Res. 3:363-377(1996).
RN [10]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=97094878; PubMed=8940112; DOI=10.1074/jbc.271.49.31145;
RA Arn E.A., Abelson J.N.;
RT "The 2'-5' RNA ligase of Escherichia coli. Purification, cloning, and
RT genomic disruption.";
RL J. Biol. Chem. 271:31145-31153(1996).
RN [11]

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RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=97061202; PubMed=8905232; DOI=10.1093/dnares/3.3.137;
RA Oshima T., Aiba H., Baba T., Fujita K., Hayashi K., Honjo A.,
RA Ikemoto K., Inada T., Itoh T., Kajihara M., Kanai K., Kashimoto K.,
RA Kimura S., Kitagawa M., Makino K., Masuda S., Miki T., Mizobuchi K.,
RA Mori H., Motomura K., Nakamura Y., Nashimoto H., Nishio Y., Saito N.,
RA Sampei G., Seki Y., Tagami H., Takemoto K., Wada C., Yamamoto Y.,
RA Yano M., Horiuchi T.;
RT "A 718-kb DNA sequence of the Escherichia coli K-12 genome
RT corresponding to the 12.7-28.0 min region on the linkage map.";
RL DNA Res. 3:137-155(1996).
RN [12]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=94261430; PubMed=8202364;
RA Fujita N., Mori H., Yura T., Ishihama A.;
RT "Systematic sequencing of the Escherichia coli genome: analysis of the
RT 2.4-4.1 min (110,917-193,643 bp) region.";
RL Nucleic Acids Res. 22:1637-1639(1994).
RN [13]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=94240115; PubMed=8183897;
RA Janosi L., Shimizu I., Kaji A.;
RT "Ribosome recycling factor (ribosome releasing factor) is essential
RT for bacterial growth.";
RL Proc. Natl. Acad. Sci. U.S.A. 91:4249-4253(1994).
RN [14]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=94124004; PubMed=7904973; DOI=10.1016/0378-1119(93)90470-N;
RA Allikmets R., Gerrard B.C., Court D., Dean M.C.;
RT "Cloning and organization of the abc and mdl genes of Escherichia
RT coli: relationship to eukaryotic multidrug resistance.";
RL Gene 136:231-236(1993).
RN [15]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=94018640; PubMed=8412694;
RA van Heeswijk W.C., Rabenberg M., Westerhoff H.V., Kahn D.D.;
RT "The genes of the glutamine synthetase adenylylation cascade are not
RT regulated by nitrogen in Escherichia coli.";
RL Mol. Microbiol. 9:443-458(1993).
RN [16]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=93259920; PubMed=8387990;
RA Zhao S., Sandt C.H., Feulner G., Vlazny D.A., Gray J.A., Hill C.W.;
RT "Rhs elements of Escherichia coli K-12: complex composites of shared
RT and unique components that have different evolutionary histories.";
RL J. Bacteriol. 175:2799-2808(1993).
RN [17]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=93123180; PubMed=8419307;
RA Yamada M., Asaoka S., Saier M.H. Jr., Yamada Y.;
RT "Characterization of the gcd gene from Escherichia coli K-12 W3110 and
RT regulation of its expression.";
RL J. Bacteriol. 175:568-571(1993).

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RN [18]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=93116053; PubMed=1474579;
RA Cormack R.S., Mackie G.A.;
RT "Structural requirements for the processing of Escherichia coli 5 S
RT ribosomal RNA by RNase E in vitro.";
RL J. Mol. Biol. 228:1078-1090(1992).
RN [19]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=93094132; PubMed=1459951;
RA Gervais F.G., Drapeau G.R.;
RT "Identification, cloning, and characterization of rcsF, a new
RT regulator gene for exopolysaccharide synthesis that suppresses the
RT division mutation ftsZ84 in Escherichia coli K-12.";
RL J. Bacteriol. 174:8016-8022(1992).
RN [20]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=93077430; PubMed=1447125;
RA Yamanaka K., Ogura T., Niki H., Hiraga S.;
RT "Identification and characterization of the smbA gene, a suppressor of
RT the mukB null mutant of Escherichia coli.";
RL J. Bacteriol. 174:7517-7526(1992).
RN [21]
RP NUCLEOTIDE SEQUENCE.
RC STRAIN=K-12;
RX MEDLINE=93011013; PubMed=1396599;
RA Condon C., Philips J., Fu Z.Y., Squires C., Squires C.L.;

Alignment Scores:

Pred. No.:	1.59e-168	Length:	464
Score:	2359.00	Matches:	464
Percent Similarity:	100.0%	Conservative:	0
Best Local Similarity:	100.0%	Mismatches:	0
Query Match:	89.1%	Indels:	0
DB:	2	Gaps:	0

US-10-812-315A-3 (1-1446) x Q2M9Q0_ECOLI (1-464)

Qy	33	ATGCCTGACGCTAAAAACAGGGGCGGTCAAACAAGGCAATGACGTTTTTCGTCTGCTTC	92
Db	1	MetProAspAlaLysLysGlnGlyArgSerAsnLysAlaMetThrPhePheValCysPhe	20
Qy	93	CTTGCCGCTCTGGCGGGATTACTCTTTGGCCTGGATATCGGTGTAATTGCTGGCGCACTG	152
Db	21	LeuAlaAlaLeuAlaGlyLeuLeuPheGlyLeuAspIleGlyValIleAlaGlyAlaLeu	40
Qy	153	CCGTTTATTGCAGATGAATTCCAGATTACTTCGCACACGCAAGAATGGGTCGTAAGCTCC	212
Db	41	ProPheIleAlaAspGluPheGlnIleThrSerHisThrGlnGluTrpValValSerSer	60
Qy	213	ATGATGTTTCGGTGCAGTCGGTGCAGCGGCTGGCTCTCCTTTAAACTCGGG	272
Db	61	MetMetPheGlyAlaAlaValGlyAlaValGlySerGlyTrpLeuSerPheLysLeuGly	80
Qy	273	CGCAAAAAGAGCCTGATGATCGGCGCAATTTGTTTGTTCGCGGTCGCTGTTCTCTGCG	332
Db	81	ArgLysLysSerLeuMetIleGlyAlaIleLeuPheValAlaGlySerLeuPheSerAla	100

Qy	333	GCTGCGCCAAACGTTGAAGTACTGATTCTTTCCGCGTTCTACTGGGGCTGGCGGTGGGT	392
Db	101	AlaAlaProAsnValGluValLeuIleLeuSerArgValLeuLeuGlyLeuAlaValGly	120
Qy	393	GTGGCCTCTTATACCGCACCGCTGTACCTCTCTGAAATTGCGCCGAAAAAATTCGTGGC	452
Db	121	ValAlaSerTyrThrAlaProLeuTyrLeuSerGluIleAlaProGluLysIleArgGly	140
Qy	453	AGTATGATCTCGATGTATCAGTTGATGATCACTATCGGGATCCTCGGTGCTTATCTTTCT	512
Db	141	SerMetIleSerMetTyrGlnLeuMetIleThrIleGlyIleLeuGlyAlaTyrLeuSer	160
Qy	513	GATACCGCCTTCAGCTACACCGGTGCATGGCGCTGGATGCTGGGTGTGATTATCATCCCG	572
Db	161	AspThrAlaPheSerTyrThrGlyAlaTrpArgTrpMetLeuGlyValIleIleIlePro	180
Qy	573	GCAATTTTGCTGCTGATTGGTGTCTTCTTCTGCCAGACAGCCACGTTGGTTTGCCGCC	632
Db	181	AlaIleLeuLeuLeuIleGlyValPhePheLeuProAspSerProArgTrpPheAlaAla	200
Qy	633	AAACGCCGTTTTGTGTATGCCGAACGCGTGCTGCTACGCCTGCGTGACACCAGCGCGGAA	692
Db	201	LysArgArgPheValAspAlaGluArgValLeuLeuArgLeuArgAspThrSerAlaGlu	220
Qy	693	GCGAAACGCGAACTGGATGAAATCCGTGAAAGTTTGCAGGTTAAACAGAGTGGCTGGGCG	752
Db	221	AlaLysArgGluLeuAspGluIleArgGluSerLeuGlnValLysGlnSerGlyTrpAla	240
Qy	753	CTGTTTAAAGAGAACAGCAACTTCCGCCGCGCGGTGTTCTTGCGTACTGTTGCAGGTA	812
Db	241	LeuPheLysGluAsnSerAsnPheArgArgAlaValPheLeuGlyValLeuLeuGlnVal	260
Qy	813	ATGCAGCAATTCACCGGGATGAACGTCATCATGTATTACGCGCCGAAAATCTTCGAACTG	872
Db	261	MetGlnGlnPheThrGlyMetAsnValIleMetTyrTyrAlaProLysIlePheGluLeu	280
Qy	873	GCGGGTTATACCAACACTACCGAGCAAATGTGGGGGACCGTGATTGTCGGCCTGACCAAC	932
Db	281	AlaGlyTyrThrAsnThrThrGluGlnMetTrpGlyThrValIleValGlyLeuThrAsn	300
Qy	933	GTACTTGCCACCTTTATCGCAATCGGCCTGTGTGACCGCTGGGGACGTAAACCAACGCTA	992
Db	301	ValLeuAlaThrPheIleAlaIleGlyLeuValAspArgTrpGlyArgLysProThrLeu	320
Qy	993	ACGCTGGGCTTCTGGTGATGGCTGCTGGCATGGGCGTACTCGGTACAATGATGCATATC	1052
Db	321	ThrLeuGlyPheLeuValMetAlaAlaGlyMetGlyValLeuGlyThrMetMetHisIle	340
Qy	1053	GGTATTCACTCTCCGTCCGCGCAGTATTTCCGCATCGCCATGCTGCTGATGTTTATTGTC	1112
Db	341	GlyIleHisSerProSerAlaGlnTyrPheAlaIleAlaMetLeuLeuMetPheIleVal	360
Qy	1113	GGTTTTGCCATGAGTGCCGGTCCGCTGATTGGGTACTGTGCTCCGAAATTCAGCCGCTG	1172
Db	361	GlyPheAlaMetSerAlaGlyProLeuIleTrpValLeuCysSerGluIleGlnProLeu	380
Qy	1173	AAAGGCCGCGATTTTGGCATCACCTGCTCCACTGCCACCAACTGGATTGCCAACATGATC	1232
Db	381	LysGlyArgAspPheGlyIleThrCysSerThrAlaThrAsnTrpIleAlaAsnMetIle	400

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Qy 1233 GTTGGCGCAACGTTCTGACCATGCTCAACACGCTGGGTAACGCCAACACCTTCTGGGTG 1292
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 Db 401 ValGlyAlaThrPheLeuThrMetLeuAsnThrLeuGlyAsnAlaAsnThrPheTrpVal 420

Qy 1293 TATGCGGCTCTGAACGTACTGTTTATCCTGCTGACATTGTGGCTGGTACCGGAAACCAA 1352
 |||
 Db 421 TyrAlaAlaLeuAsnValLeuPheIleLeuLeuThrLeuTrpLeuValProGluThrLys 440

Qy 1353 CACGTTTTCGCTGGAACATATTGAACGTAATCTGATGAAAGGTCGTAAACTGCGCGAAATA 1412
 |||
 Db 441 HisValSerLeuGluHisIleGluArgAsnLeuMetLysGlyArgLysLeuArgGluIle 460

Qy 1413 GGCGCTCACGAT 1424
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 Db 461 GlyAlaHisAsp 464

1: U00096. Reports Escherichia coli ...[gi:48994873] Links

CommentFeaturesLOCUS U00096 4639675 bp DNA circular BCT 08-SEP-2006

DEFINITION Escherichia coli K12 MG1655, complete genome.

ACCESSION U00096 AE000111-AE000510

VERSION U00096.2 GI:48994873

KEYWORDS .

SOURCE Escherichia coli K12

ORGANISM Escherichia coli K12

Bacteria; Proteobacteria; Gammaproteobacteria; Enterobacteriales;
 Enterobacteriaceae; Escherichia.

REFERENCE 1 (bases 1 to 4639675)

AUTHORS **Blattner**,F.R., Plunkett,G., Bloch,C.A., Perna,N.T., Burland,V.,
 Riley,M., Collado-Vides,J., Glasner,J.D., Rode,C.K., Mayhew,G.F.,
 Gregor,J., Davis,N.W., Kirkpatrick,H.A., Goeden,M.A., Rose,D.J.,
 Mau,B. and Shao,Y.

TITLE The complete genome sequence of Escherichia coli K-12

JOURNAL Science 277 (5331), 1453-1474 (1997)

PUBMED 9278503

REFERENCE 2 (bases 1 to 4639675)

AUTHORS Riley,M., Abe,T., Arnaud,M.B., Berlyn,M.K., Blattner,F.R.,
 Chaudhuri,R.R., Glasner,J.D., Horiuchi,T., Keseler,I.M., Kosuge,T.,
 Mori,H., Perna,N.T., Plunkett,G. III, Rudd,K.E., Serres,M.H.,
 Thomas,G.H., Thomson,N.R., Wishart,D. and Wanner,B.L.

TITLE Escherichia coli K-12: a cooperatively developed annotation
 snapshot--2005

JOURNAL (er) Nucleic Acids Res. 34 (1), 1-9 (2006)

PUBMED 16397293

REFERENCE 3 (bases 1 to 4639675)

AUTHORS Arnaud,M., Berlyn,M.K.B., Blattner,F.R., Galperin,M.Y.,
 Glasner,J.D., Horiuchi,T., Kosuge,T., Mori,H., Perna,N.T.,
 Plunkett,G. III, Riley,M., Rudd,K.E., Serres,M.H., Thomas,G.H. and
 Wanner,B.L.

TITLE Workshop on Annotation of Escherichia coli K-12

JOURNAL Unpublished

REMARK Woods Hole, Mass., on 14-18 November 2003 (sequence corrections)

REFERENCE 4 (bases 1 to 4639675)

AUTHORS Glasner,J.D., Perna,N.T., Plunkett,G. III, Anderson,B.D.,
 Bockhorst,J., Hu,J.C., Riley,M., Rudd,K.E. and Serres,M.H.

TITLE ASAP: Escherichia coli K-12 strain MG1655 version m56

Art Unit: 1656

JOURNAL Unpublished
REMARK ASAP download 10 June 2004 (annotation updates)
REFERENCE 5 (bases 1 to 4639675)
AUTHORS Hayashi,K., Morooka,N., Mori,H. and Horiuchi,T.
TITLE A more accurate sequence comparison between genomes of Escherichia coli K12 W3110 and MG1655 strains

JOURNAL Unpublished
REMARK GenBank accessions AG613214 to AG613378 (sequence corrections)
REFERENCE 6 (bases 1 to 4639675)
AUTHORS Perna,N.T.
TITLE Escherichia coli K-12 MG1655 yqiK-rfaE intergenic region, genomic sequence correction

JOURNAL Unpublished
REMARK GenBank accession AY605712 (sequence corrections)
REFERENCE 7 (bases 1 to 4639675)
AUTHORS Rudd,K.E.
TITLE A manual approach to accurate translation start site annotation: an E. coli K-12 case study

JOURNAL Unpublished
REFERENCE 8 (bases 1 to 4639675)
AUTHORS Blattner,F.R. and Plunkett,G. III.
TITLE Direct Submission
JOURNAL Submitted (16-JAN-1997) Laboratory of Genetics, University of Wisconsin, 425G Henry Mall, Madison, WI 53706-1580, USA

REFERENCE 9 (bases 1 to 4639675)
AUTHORS Blattner,F.R. and Plunkett,G. III.
TITLE Direct Submission
JOURNAL Submitted (02-SEP-1997) Laboratory of Genetics, University of Wisconsin, 425G Henry Mall, Madison, WI 53706-1580, USA

REFERENCE 10 (bases 1 to 4639675)
AUTHORS Plunkett,G. III.
TITLE Direct Submission
JOURNAL Submitted (13-OCT-1998) Laboratory of Genetics, University of Wisconsin, 425G Henry Mall, Madison, WI 53706-1580, USA

REFERENCE 11 (bases 1 to 4639675)
AUTHORS Plunkett,G. III.
TITLE Direct Submission
JOURNAL Submitted (10-JUN-2004) Laboratory of Genetics, University of Wisconsin, 425G Henry Mall, Madison, WI 53706-1580, USA

REMARK Sequence update by submitter
REFERENCE 12 (bases 1 to 4639675)
AUTHORS Plunkett,G. III.
TITLE Direct Submission
JOURNAL Submitted (07-FEB-2006) Laboratory of Genetics, University of Wisconsin, 425G Henry Mall, Madison, WI 53706-1580, USA

REMARK Protein updates by submitter
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ORIGIN

//

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Sequences producing significant alignments:	Score (Bits)	E Value
gi 85674274 dbj AP009048.1 Escherichia coli W3110 DNA, complete	2841	0.0
gi 48994873 gb U00096.2 Escherichia coli K12 MG1655, complete g	2841	0.0
gi 882431 gb U28377.1 ECU28377 Escherichia coli K-12 genome; app	2841	0.0
gi 81244029 gb CP000036.1 Shigella boydii Sb227, complete genom	2785	0.0
gi 73854091 gb CP000038.1 Shigella sonnei Ss046, complete genom	2769	0.0
gi 81239530 gb CP000034.1 Shigella dysenteriae Sd197, complete	2714	0.0
gi 110341805 gb CP000247.1 Escherichia coli 536, complete genom	2698	0.0
gi 26111730 gb AE014075.1 Escherichia coli CFT073, complete gen	2690	0.0
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Features in this part of subject sequence:

D-galactose transporter

Score = 2841 bits (1433), Expect = 0.0
Identities = 1433/1433 (100%), Gaps = 0/1433 (0%)
Strand=Plus/Plus

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